Kidney International, Vol. 37 (1990), pp. 1402–1408

RAPID COMMUNICATIONS

Tubuloglomerular feedback dependence of autoregulation in rat juxtamedullary afferent arterioles

LEON C. MOORE and DANIEL CASELLAS

Department of Physiology and Biophysics, State University of New York, Stony Brook, New York, USA and Groupe Rein et Hypertension, St. Charles Hospital, Montpellier, France

Tubuloglomerular feedback dependence of autoregulation in rat juxtamedullary afferent arterioles. Experiments were performed in blood-perfused juxtamedullary nephrons in vitro to evaluate the tubuloglomerular feedback (TGF) dependence of autoregulatory vasoconstriction in mid-to-late (mAA) and juxtaglomerular (jAA) afferent arterioles. Videometric measurements were made of perfusion pressure (PP) dependent changes in lumen diameter of superficial vessels before and after acute inhibition of the TGF mechanism by direct microinfusion of 0.1 mM furosemide solution into the macula densa (MD) segment. When PP was raised from 60 to 123 ± 7 mm Hg in seven vessels, jAA diameter decreased by 29 ± 3% (SEM, $N = 7$). During furosemide infusion with the same change in PP, jAA diameter decreased only 7 ± 2%. After calcium channel blockade with 1 micromolar nimodipine, jAA lumen diameter increased by 21 ± 7%. A similar pattern of responses was observed in eight jAA where TGF was inhibited with an oil block at the MD. mAA autoregulatory responses were also blunted by TGF inhibition. Raising PP from 60 to 120 mm Hg resulted in 15 ± 2% and 7 ± 2% decreases in mAA luminal diameter before and after TGF inhibition. These results demonstrate that the autoregulatory responses in mid- and juxtaglomerular afferent arteriolar segments are mediated by both TGF and a TGF-independent myogenic mechanism.

Autoregulation of renal blood flow and glomerular filtration rate is generally recognized to be the consequence of adjustments in preglomerular vascular resistance [1–9]. Nevertheless, the relative contributions of the different arterial segments and the mechanisms that underlie renal autoregulation remain controversial. Strong autoregulatory vasoconstriction has been reported in late [2, 3, 7] and in the terminal juxtaglomerular segment of the afferent arteriole (jAA) in blood-perfused juxtamedullary (JM) nephrons [1]. Conversely, Steinhausen et al [9] have found this segment to be unaffected by perfusion pressure in split hydronephrotic rat kidneys. Edwards [10] found a similar lack of autoregulatory responses in isolated rabbit late AA. As these two preparations lack an intact tubular system, one possible reason for these differences in behavior is the loss of tubuloglomerular feedback (TGF), which many studies have shown to be an important mediator of renal autoregulation [5–8]. Support for this explanation has been provided by Sanchez-Ferrer, Roman and Harder [7], who demonstrated that AA autoregulatory responses in juxtamedullary (JM) nephrons in vitro were abolished by acute TGF inhibition.

The relative contribution of TGF to renal autoregulation has also recently been questioned. Several studies in rat have demonstrated a significant TGF-independent, myogenic component of renal autoregulation [2, 4, 6, 8, 9, 11–13]. In contrast, Sakai, Hallman and Marsh [14] have argued against such a myogenic mechanism, based on their inability to identify any significant regulatory mechanism with a time response different than that of the TGF mechanism, although others have reached different conclusions with the same techniques [11]. Further, evidence of strong autoregulation in the presence of constant maximal TGF stimulation in single nephrons has been documented [12, 13]. On the basis of these studies, Davis, Kawata and Häberle [12] have concluded that TGF is not an essential element of renal autoregulation.

We recently demonstrated furosemide inhibitable TGF responses in the jAA and strong autoregulatory responses in the AA in blood-perfused JM nephrons in vitro [1], a preparation that provides direct experimental access to the macula densa (MD) and the entire AA. The goals of the present study of JM AA in vitro were to determine: 1) the extent to which the autoregulatory responses in the jAA and mAA are mediated by the TGF mechanism, and 2) the magnitude of the TGF-independent myogenic autoregulatory responses in the AA. The results support the concept that renal autoregulation is mediated by both TGF and myogenic mechanisms.

Methods

Studies were conducted in 21 kidneys from male Sprague-Dawley rats (Charles River, Montpellier, France), weighing 250 to 350 g and anesthetized with an i.p. injection of 50 mg/kg sodium pentobarbital. The techniques used for the perfusion of JM nephrons in vitro have been described in detail previously [15]. Briefly, the left kidney was acutely denervated with phenol in situ and perfusion established via an aortic catheter with a Krebs-bicarbonate-Ringer (KBR) solution equilibrated with 93% O_2/7% CO_2 to pH 7.4. The KBR perfusate was a normal Ringer containing 4% dialyzed bovine serum albumin (Fraction V, Sigma Chemical Corp., St. Louis, Missouri, USA) and 5 mM Hepes buffer. The perfusate also contained a mixture of L-amino acids [1]. The kidney was removed, sectioned longitudinally, and the papilla reflected upwards, exposing the inner surface of the cortex. The overlying pelvic mucosa was re-

Received for publication December 18, 1989
and in revised form March 7, 1990
Accepted for publication March 13, 1990

moved, large veins were opened, and all major arteries supplying the rest of the kidney were ligated. During dissection, the kidney was perfused with KBR perfusate at room temperature. During measurements, the kidney was perfused with a blood solution prepared from fresh rat blood. The red cells were separated, washed, and resuspended in filtered (0.45 μ) KBR-albumin solution with 6% albumin to a hematocrit of ~30%. The perfusion system was as described previously [15], except for the addition of a glass bead filter, which we have found to minimize clogging of glomerular capillaries. The preparation was superfused at high rates (~3 ml/min) with a warmed (37°C) KBR solution containing 1% albumin and 5 mM Hepes buffer with pH 7.4. The preparation was mounted in a small perfusion chamber placed on a microscope stage. After preparation, ~10% of the kidney remains perfused. In this area, afferent and efferent arterioles, the glomerulus, and the MD segment are visible on the surface.

Shadow-cast images of superficial structures were obtained as described previously [1]. A fixed-stage compound microscope (Leitz LaborLux D-FS, Heerbrugg, Switzerland) with a long-working distance objective (Leitz, NPL Fluotar L 25/0.35) was used. The surface of the preparation was illuminated with low-angle (~30°) incident light via a small (0.8 mm) fiber optic light pipe placed under the superfusate layer. Images were displayed and recorded with a video system consisting of a 1.25× camera adaptor, black and white CCD camera (Cohu model 4710), video monitor, and a 3/4 inch video recorder (SONY VO-5800PS, Tokyo, Japan). The images were contrast enhanced with an analog instrument with dimensional measuring capability (For-A, IV550). The final magnification on the video screen was 1300×. Vessel diameters were measured directly on the video screen using the electronic video caliper system; all values were verified later from video tape recordings.

Superficial JM nephrons with visible jAA and cTAL were selected for study. The 21 kidneys were divided into three groups. In the first group of seven nephrons, the effect on jAA autoregulation of TGF inhibition by furosemide microinfusion into the MD was evaluated. First, control measurement of jAA lumen diameter were made at 60 and ~120 mm Hg perfusion pressure. A lower perfusion pressure (PP) limit of 60 mm Hg was chosen because JM nephrons autoregulate over a lower pressure range than superficial cortical nephrons [16], and TGF in JM nephrons is quiescent at 60 mm Hg [1]. Luminal diameters were measured three to five minutes after the change in PP. The segment of cortical thick ascending limb (cTAL) containing the MD then was located by visual inspection. The cTAL has a smaller diameter than proximal tubules, it makes contact with the jAA at the vascular pole, and it is closely juxtaposed with the efferent arteriole (EA) in the region between the glomerulus and the cortico-medullary boundary [1]. A micromanipulator (Leitz) was then used to puncture the cTAL with a double-sharpened micropipette (4 μ O.D.) filled with stained (0.1% FD&C green) Ringers solution containing 0.1 mM furosemide, an amount sufficient to completely block TGF [17]. In six nephrons, the puncture site was ~150 μ upstream from the MD region; in 1 nephron it was ~20 μ downstream. The micropipette was connected to a Landis-type mercury manometer, which was used to change pipette back pressure and, thereby, fluid ejection out of the pipette. After puncture, pipette back pressure was briefly increased to ~20 mm Hg to inject a small bolus of stained fluid into the cTAL to verify tubular flow and to map the cTAL and MD. Pipette back pressure was then increased to ~40 mm Hg, resulting in a brisk, continuous infusion of furosemide solution. Although the rate of infusion was not measured, tubular distension and the presence of stained fluid at the MD was evident in every nephron. Also, we previously demonstrated that this microinjection technique is sufficient to completely inhibit TGF [1]. Lumen diameter of the jAA was remeasured at 60 mm Hg PP ~ one to two min after the beginning of the furosemide microinjection. PP was then raised slowly to 120 mm Hg while carefully monitoring the position of the pipette to maintain the furosemide injection, and vessel diameter measured. PP was then lowered to 60 mm Hg, and lumen diameter was remeasured. The EA and mAA segment could not be visualized in this series, as the field of vision could not be moved while a pipet was in the tubule. Finally, to characterize the behavior of the vessels in the absence of calcium-dependent smooth muscle contraction, jAA autoregulation was remeasured in six of the seven vessels after calcium channel blockade, accomplished by adding 1 μM nimodipine (Bayer) to the superfusate. The supply of blood perfusate was inadequate in one experiment to evaluate calcium channel blockade. jAA diameter was remeasured at PP of 60 and 120 mm Hg beginning 30 minutes after the start of nimodipine superfusion.

In the second series conducted in eight rats, TGF blockade was accomplished by placing an immobile oil block in the MD segment. In this series, autoregulation measurements were made in both jAA and mAA segments. In four of the eight nephrons, it was also possible to measure the autoregulatory responses of the early EA. PP was increased from 60 to 140 mm Hg in 20 mm Hg steps and then decreased back to 60 mm Hg in 40 mm Hg steps. Vessel diameter was measured at each PP step. Nephron flow was then verified by the injection of a small bolus of stained Ringers solution into the cTAL with a micropipette and observing its washout. The TGF system was then inhibited by filling the MD region with stained castor oil and establishing an upstream vent. The autoregulation measurements were then repeated. Finally, autoregulatory responses were measured a third time in five of the eight nephrons 30 minutes after adding 1 μM nimodipine to the superfusate.

The third group of six rats was used for time-control studies conducted to verify the stability of autoregulation over the time required to complete the oil blockade protocol (excluding the measurements during nimodipine). Two measurements of autoregulatory responses, using the same PP steps as in the oil blockade series, were made in jAA, mAA, and EA (N = 5) segments separated by an interval of ~42 minutes, which was the average time required to verify tubular flow and achieve TGF blockade. The result was that corresponding measurements were separated by ~120 minutes. The effect of nimodipine was not evaluated in these kidneys.

Whenever significant vasomotion was present (>1 μ change in luminal diameter), the average of the minimum and maximum values were recorded. The individual diameter measurements were averaged at each level of PP for each vessel. The autoregulatory responses in the time-control and oil block series were analyzed by calculating the slope by linear regression of the autoregulatory pressure-diameter relationship measured in each individual vessel. Statistical analysis of the differences

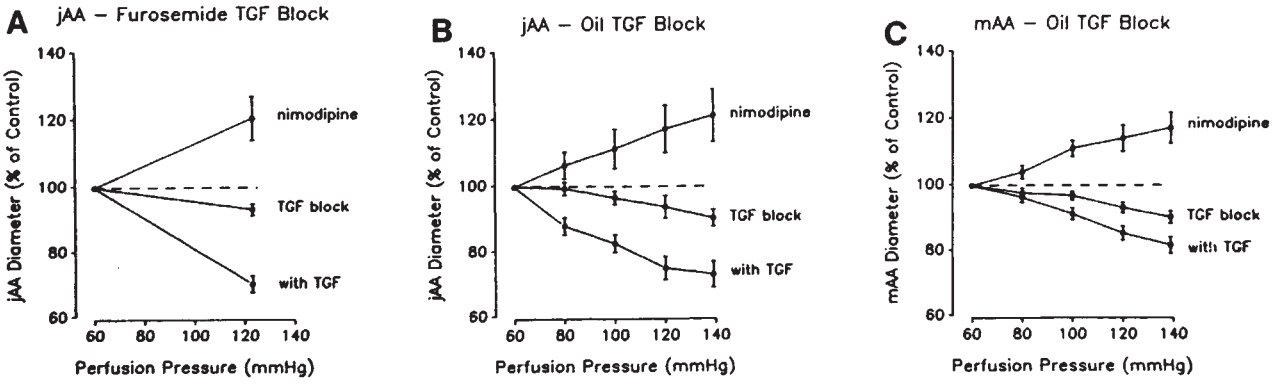

Fig. 1. *Autoregulatory responses in juxtamedullary afferent arterioles.* **A.** Responses (mean ± SEM) of the juxtaglomerular segment of the AA (jAA) before and after inhibition of the TGF system by the direct infusion of 0.1 mM furosemide solution into the macula densa region ($N = 7$), and after calcium channel blockade with 1 μM nimodipine ($N = 6$). **B.** Autoregulatory responses in 8 jAA with TGF intact, during TGF blockade by injection of oil into the macula densa segment, and after calcium channel blockade with nimodipine ($N = 5$). **C.** Autoregulatory responses in mid-afferent (mAA) segments of the same 8 vessels as in the middle panel.

between autoregulatory pressure-diameter curves before and after calcium channel blockade or TGF inhibition was accomplished by unpaired comparison of the mean response slopes, using Student's *t*-test with a $P < 0.05$ considered to be significant. All values are presented as mean ± SEM.

Results

The mean lengths of the entire AA and jAA segments were 489 ± 44 and 52 ± 2 μ. The jAA was identified on the basis of increased wall thickness in the jAA segment as compared to the mAA (6.4 ± 0.2 vs. 4.6 ± 0.2 μ, respectively). In the jAA, the lumen also narrowed by an average of 33 ± 3% at 60 mm Hg PP. These features coincide with the presence of renin-positive granulated cells in the jAA [18, 19]. The jAA and mAA measuring sites were 21 ± 2 and 134 ± 6 μ from the vascular pole, respectively; the EA site was 63 ± 26 μ from the exit from Bowman's capsule in the four nephrons in which it was visible. Occasional significant vasomotion was seen in 7 of the 21 nephrons.

Figure 1A shows the effect of intraluminal furosemide and nimodipine treatment on jAA autoregulatory responses in seven vessels. At 60 mm Hg, jAA diameter was 12.6 ± 1.3 μ. Increasing PP to 123 ± 7 mm Hg led to a significant (32 ± 4%) reduction in vessel caliber to 8.4 ± 1.0 μ, corresponding to a mean response slope of −0.067 ± 0.011 μ/mm Hg. Continuous injection of 0.1 mM furosemide into the MD had no significant effect at 60 mm Hg on jAA diameter (12.5 ± 1.1 μ). However, it did significantly reduce autoregulatory vasoconstriction to 8 ± 2%. At high PP, jAA diameter was 11.4 ± 1.0 μ during furosemide, corresponding to a reduced but significant response slope of −0.020 ± 0.004 μ/mm Hg. Six of the seven vessels were treated with 1 μM nimodipine, which increased jAA diameter at 60 mm Hg PP significantly by 3.3 ± 1.1 μ (paired *t*-test, $N = 6$). With increased PP, the jAA passively dilated by 21 ± 6%, from 15.4 ± 2.0 at 60 mm Hg to 18.7 ± 2.7 μ at 123 ± 7 mm Hg. The mean response slope was 0.053 ± 0.018 μ/mm Hg. These results demonstrate that in the jAA active autoregulatory vasoconstriction is highly dependent upon TGF. The results also reveal a substantial myogenic component of segmental vascular tone, as calcium channel inhibition produced significant PP-dependent vasodilation.

The micrographs in Figure 2 illustrate the effect of TGF inhibition on jAA autoregulatory responses. In this nephron, the MD region is located just to the left of the jAA, and it appears black when filled with stained castor oil. The pattern of autoregulatory responses in jAA diameter are similar to those observed during furosemide blockade: substantial loss of autoregulatory vasoconstriction during TGF inhibition.

The results of TGF inhibition by oil blockade in eight nephrons are summarized in Figure 1 (B and C). In the jAA, mean diameter was 15.3 ± 1.8 μ at 60 mm Hg, which decreased significantly by 27 ± 4% to 11.4 ± 1.6 μ at 140 mm Hg with an average slope of −0.049 ± 0.006 μ/mm Hg. Without TGF, jAA diameter was unchanged at 60 mm Hg (15.3 ± 2.0) but autoregulatory vasoconstriction was significantly reduced at 140 mm Hg. Lumen diameter decreased by only 10 ± 3% to 13.8 ± 1.8 μ with a mean response slope of −0.020 ± 0.006 μ/mm Hg. While greatly reduced by TGF inhibition, the residual autoregulatory vasoconstriction remained significant (paired *t*-test), although the PP diameter curve seemed to be markedly shifted toward higher levels of PP. The effects of nimodipine were examined in five of the eight nephrons. At 60 mm Hg, jAA diameter was significantly larger than in control, averaging 18.1 ± 2.2 μ ($N = 5$, paired *t*-test). With increased PP, the jAA passively dilated with a significant mean slope of 0.042 ± 0.010 μ/mm Hg. At 140 mm Hg PP, mean jAA diameter was 21.5 ± 1.9 μ, corresponding to a 21 ± 8% dilation from its value at 60 mm Hg.

In the mAA segment, mean diameters at 60 and 140 mm Hg were 24.0 ± 0.8 and 19.8 ± 1.0 μ, corresponding to a significant 18.1 ± 2.5% vasoconstriction with a mean response slope of −0.057 ± 0.007 μ/mm Hg. Placing an oil block at the MD had no effect at 60 mm Hg on mAA diameter (23.8 ± 0.8 μ). TGF inhibition significantly reduced mAA autoregulatory vasoconstriction. mAA diameter was 21.6 ± 1.1 μ at 140 mm Hg, corresponding to a residual, TGF independent vasoconstriction of 9.6 ± 1.9% with a response slope of −0.027 ± 0.005 μ/mm Hg. Although the difference in the responses before and after

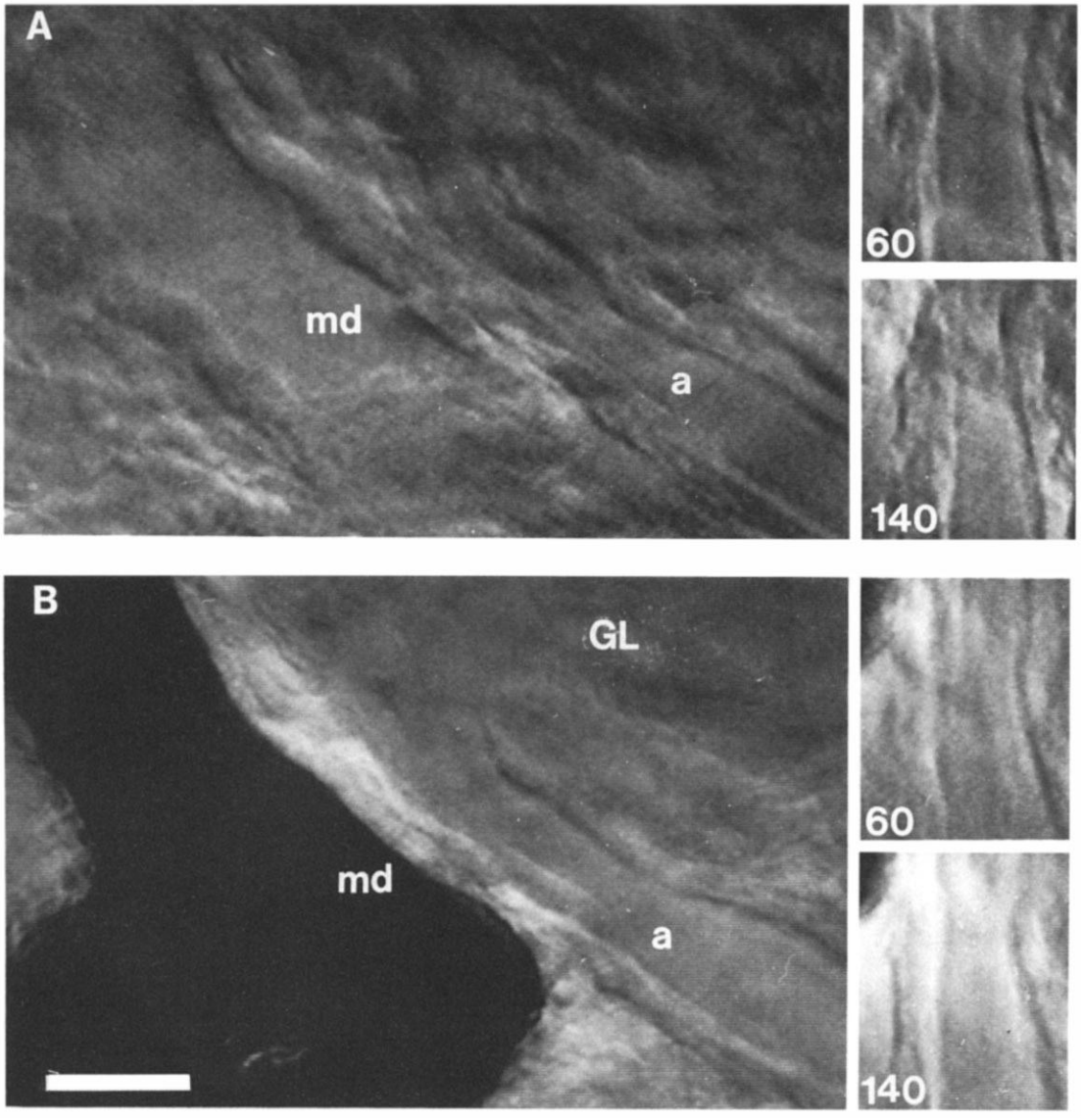

Fig. 2. *Effect of TGF inhibition on autoregulation in the jAA.* **A.** The large panel shows the lumen of the jAA (a) and the macula densa segment (md) during control measurements at 100 mm Hg perfusion pressure (PP). On the right are views of the same jAA at PP of 60 and 140 mm Hg. The camera was rotated and the location is approximately the same as the symbol a on the large panel. **B.** The same nephron showing the macula densa region and surrounding cTAL filled with stained castor oil. The glomerular tuft is indicated by GL. The photos on the right show the attenuation of jAA autoregulatory vasoconstriction after TGF blockade. The large photograph in B, which was selected to match the orientation of the corresponding photograph in A, was taken after nimodipine treatment. The small panels were taken after TGF blockade but prior to nimodipine superfusion. The magnification is the same in all panels and the bar in B corresponds to 25 μ. The photographs are of the video screen during playback of videotape recordings.

TGF inhibition appears to be more modest than in the jAA segment, it is highly significant ($P < 0.001$, paired t-test). Like the jAA, TGF inhibition appears to have shifted the PP-diameter response curve towards higher levels of PP. After calcium channel blockade in five of the eight nephrons, mAA diameter was slightly but not significantly larger at 60 mm Hg (25.1 ± 1.1 μ). As PP was increased to 140 mm Hg, the mAA segment dilated by 17 ± 5% to a diameter of 29.2 ± 1.6 μ with a mean response slope of 0.054 ± 0.017 μ/mm Hg.

The early EA was visible in four of the eight nephrons studied in the oil block series. Mean diameter was 23.8 ± 3.1 μ at 60 mm Hg, and segment diameter was unaffected by increased PP, with an average response slope of 0.010 ± 0.004 μ/mm Hg. A similar insensitivity to PP was observed during TGF inhibition, with a mean diameter of 23.8 ± 2.8 μ at 60 mm Hg and a response slope of 0.005 ± 0.004 μ/mm Hg. Only two EA were examined during nimodipine treatment, and neither vessel vasodilated substantially at any level of PP.

The results of the time-control studies in six nephrons are shown in Figure 3. In the jAA, the diameter at all levels of PP increased slightly after ~120 minutes, with significant mean increases of 0.6 ± 0.2 and 1.2 ± 0.3 μ at 60 and 120 mm Hg. Nevertheless, the responsiveness of the jAA, as expressed by the mean PP-diameter response slope, was unchanged (−0.056

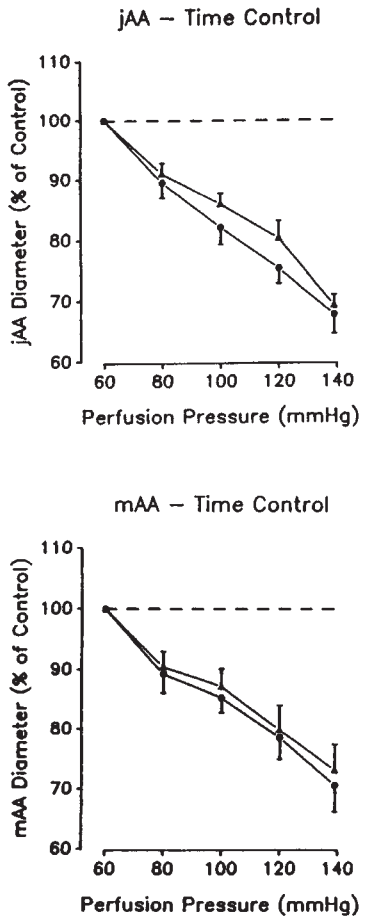

Fig. 3. *Results of time-control studies in 6 nephrons.* Symbols are: (●——●) initial; (▲——▲) 120 min. Autoregulatory responses were remeasured after 120 min, the approximate period of time required to complete the TGF oil blockade studies. Values are mean ± SEM.

± 0.010 vs. −0.052 ± 0.007 μ/mm Hg). The mAA showed no tendency to dilate with time, with the largest, insignificant change (0.4 ± 0.5 μ) occurring at 140 mm Hg PP. The mean slope of the autoregulatory response curve was also unchanged (−0.075 ± 0.010 vs. −0.072 ± 0.015 μ/mm Hg). These results indicate that the observed attenuation of autoregulation by TGF inhibition can not be attributed to a gradual loss of responsiveness of the preparation over the period of time required to make the measurements.

To explore the functional significance of these data, the relative increments in the structural component of AA segmental resistance were calculated from the changes in vessel caliber. The calculation assumes an inverse fourth power relationship between vessel radius and resistance and constant blood viscosity. The results are shown in Figure 4. In response to a doubling of PP from 60 to 120 mm Hg, jAA segmental resistance increased ~ fourfold, a large change that is similar in magnitude to the effect of TGF-stimulation on jAA resistance [1]. Conversely, mAA segmental resistance increased in proportion to the change in PP. TGF inhibition significantly decreased the resistance increment in both segments. The most pronounced effect was in the jAA, where the relative increase in resistance was reduced by ~86%, from 3.9 ± 1.2 to 1.4 ± 0.2.

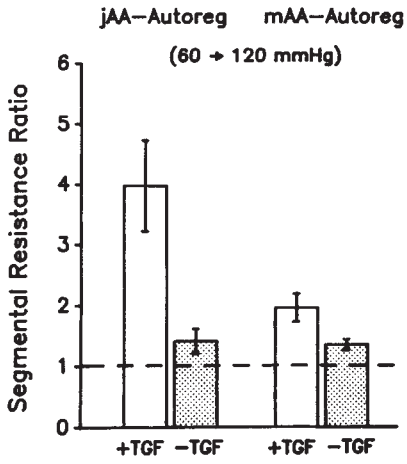

Fig. 4. *Estimated increases in the structural component of segmental resistance elicited by a doubling of perfusion pressure.* The values were calculated from the 8 individual measurements made in the TGF oil block series, assuming an inverse fourth-power relationship between vessel radius and vascular resistance. The dotted line at unity indicates no change. Values are mean ± SEM.

However, the change in *total* AA resistance is likely to more closely reflect the response of the mAA, owing to the fact that the jAA constitutes only ~12% of the length of the AA. In the mAA, TGF inhibition reduced the segmental resistance ratio from 1.96 ± 0.23 to 1.35 ± 0.09, corresponding to a ~60% reduction in the autoregulatory resistance increase in this segment.

Discussion

This study provides the first direct observations of the effects of TGF inhibition in a single nephron on segmental autoregulatory responses in the associated afferent arteriole. The results demonstrate that autoregulation in the jAA is highly dependent upon TGF, with a smaller but significant TGF-independent myogenic component. Further, the results show that autoregulation in the mAA segment is also significantly influenced by TGF, with a more pronounced myogenic capability. These findings extend an earlier investigation that demonstrated strong autoregulatory and TGF responses in the jAA segment of JM nephrons [1]. In contrast, the early EA does not appear to participate in autoregulation in these nephrons, as observed previously [1]. The constancy of EA diameter, even when preglomerular autoregulatory responses are blocked and during calcium channel blockade [1, 9], suggests that the early EA merely provides a constant post-glomerular resistance. However, as these measurements were made with an artificial plasma, any EA response requiring a humoral factor in the blood, such as angiotensinogen, would be suppressed in this study.

The dependence of AA autoregulatory vasoconstriction on TGF is consistent with many studies of autoregulation in superficial nephrons [5, 8, 12, 13] and whole kidneys [14]. The results also provide a possible explanation for the lack of substantial autoregulatory vasoconstriction in late AA in hydronephrotic kidneys [9] and in AA segments in vitro [10], where the renal tubular system and, hence, TGF are absent. Indeed, the amount of autoregulatory vasoconstriction seen here during

TGF inhibition is similar in magnitude to that seen in preparations without an intact tubular system—a ~10% reduction in diameter for a ~80 to 100 mm Hg change in PP.

The results also demonstrate a substantial TGF-independent myogenic autoregulatory component in both the mAA and jAA segments, again in agreement with micropuncture studies in superficial nephrons [5, 6, 8, 12, 13] and a variety of studies in whole kidney preparations [6, 9, 11]. Further, such myogenic capacity appears to be present along the entire preglomerular JM vasculature [2, 3]. Our results also provide an estimate of the relative contribution of the two mechanisms, although this may well change under different experimental and physiological settings. Most of the PP-dependent vasoconstriction in the jAA and much of it in the mAA is dependent on TGF. However, the myogenic component seems to be of nearly equal magnitude when the total, calcium-dependent response is considered. In the absence of TGF, most of the work done by the myogenic component in the jAA appears to counteract vessel distention, so that the net vasoconstriction is small. In the mAA, the relative contribution of the myogenic mechanism is larger, but again, full compensation seems to be dependent upon the presence of an intact TGF system.

Sanchez-Ferrer et al [7] previously reported strong TGF-dependence of autoregulation in JM vessels in vitro, a finding consistent with the present study. However, our results differ substantially from theirs in two respects. First, a significant fraction of the AA they examined (41%) dilated with increased PP, whereas we observed vasoconstriction in every AA in this and an earlier study [1]. Second, they concluded that the autoregulatory responses were completely dependent upon TGF, with little evidence of a myogenic autoregulatory component. This conclusion was based on their observation that additional AA vasodilation, beyond that associated with TGF inhibition by papillectomy or adding furosemide to the perfusate, could not be elicited by removal of calcium from the perfusate or with a mixture of vasodilators. These differences are most likely related to the use of a red-cell free perfusion medium. We have found that this inhibits myogenic responsiveness, particularly in arcuate and interlobular arteries, such that all vessels are dilated and the large arteries distend with increased PP (unpublished observations). This is exactly what was observed by Sanchez-Ferrer and co-workers [7], but is opposite to the consistent observation of significant PP-dependent vasoconstriction in all preglomerular vessels in JM nephrons perfused with blood or solutions containing red cells [2, 3]. The nature of the mechanisms underlying this apparent dependence of myogenic reactivity on red cells, whether physical, such as by altering the shear stress exerted on endothelial cells, or humoral, by disturbing the levels of vasoactive substances, remains to be determined.

An interesting question arises from the observation that TGF inhibition reduced mAA autoregulation by ~60%: What is the mechanism by which a TGF response in the jAA results in enhanced vasoconstriction ~100 μ upstream? One obvious possibility is electronic conduction of a local TGF signal for vasoconstriction. Although there are numerous myoendothelial junctions between endothelial cells in the AA, gap junctions are less prevalent between smooth muscle cells [20]. In a recent study of AA from hydronephrotic mouse kidneys, Noibling and Bührle [21] found that the extent of coupling was only three to four cells. Based on these data, they suggest that the AA has characteristics similar to a multi-unit vessel, where local stimuli are not propagated over long distances along the arteriole. Although the issue of whether the AA is a single- or multi-unit vessel needs additional study, there is another possible mechanism which would act in concert with any electrotonic coupling that may exist. This second mechanism is based on the fact that a strong TGF response, localized in the jAA, could enhance mAA myogenic vasoconstriction indirectly by increasing upstream intravascular pressure. We [22] and Davis et al [12] have developed mathematical models of such intravascular-pressure-dependent amplification of myogenic tone by downstream vasoconstriction, a phenomenon we have termed *ascending myogenic autoregulation*. Our model suggests that such ascending myogenic autoregulation is sufficiently strong to account for the degree of TGF-dependency of mAA autoregulation observed in the present study. The models also predict enhanced myogenic autoregulation in single nephrons when TGF is under constant, maximal stimulation, which agrees with experimental studies [12, 13]. Although the concepts embodied in these mathematical models are of potential importance in understanding the regulation of the preglomerular circulation, direct experimental verification of many of the underlying assumptions remains to be done.

Acknowledgments

Supported by Grants from INSERM (CRE 875003) and NIDDKD (DK 26341). L.C. Moore was the recipient of a Poste Orange Fellowship from INSERM. Annie Artuso and Aija Birzgalis provided essential technical assistance. We thank P. Schuman for printing the micrographs.

Reprint requests to Leon C. Moore, Ph.D., Department of Physiology and Biophysics, SUNY Health Science Center, Stony Brook, New York 11794, USA.

References

1. Casellas D, Moore LC: Autoregulation and tubuloglomerular feedback in juxtamedullary glomerular arterioles. *Am J Physiol* 258:F660–F669, 1990
2. Casellas D, Moore LC: Distribution of autoregulatory responses in preglomerular vessel of rat juxtamedullary nephrons. (abstract) *FASEB J* 3:A541, 1989
3. Carmines PK, Veldkamp PJ, Navar LG: Responses of in vitro blood-perfused juxtamedullary nephrons to alterations in perfusion pressure, in *The Juxtaglomerular Apparatus*, edited by AEG Persson, U Boberg, Amsterdam, Elsevier Science Publishers, 1988, pp. 457–459
4. Källskog Ö, Lindbom LO, Ulfendahl HR, Wolgast M: Hydrostatic pressures within the vascular structures of the rat kidney. *Pflügers Arch* 363:205–210, 1976
5. Moore LC: Tubuloglomerular feedback and SNGFR autoregulation in the rat. *Am J Physiol* 247:(Renal, Fluid Electrol Physiol 16:)F267–F276, 1984
6. Ofstadt, Aukland K: The renal circulation, in *The Kidney: Physiology and Pathophysiology*, edited by DW Seldin, G Giebisch, New York, Raven Press, Chapter 21, 1985
7. Sanchez-Ferrer CF, Roman RJ, Harder DR: Pressure-dependent contraction of rat juxtamedullary afferent arterioles. *Circ Res* 64:790–798, 1989
8. Schnermann J, Briggs J: Function of the juxtaglomerular apparatus: Local control of glomerular hemodynamics, in *The Kidney: Physiology and Pathophysiology*, edited by DW Seldin, G Giebisch, New York, Raven Press, Chapter 28, 1985

9. STEINHAUSEN M, BLUM M, FLEMING JT, HOLZ FG, PAREKH N, WEIGMAN DL: Visualization of renal autoregulation in the split hydronephrotic kidney of rats. *Kidney Int* 35:1151–1160, 1989
10. EDWARDS RM: Segmental effects of norepinephrine and angiotensin II on isolated renal microvessels. *Am J Physiol* 244:(Renal, Fluid Electrol Physiol 13:)F526–F534, 1983
11. DANIELS FH, ARENDSHORST WJ: Autoregulation of renal blood flow is not impaired by abolition of tubuloglomerular feedback. (abstract) *FASEB J* 3:A1016, 1989
12. DAVIS JM, KAWATA T, HÄBERLE DA: The influence of renal autoregulation on TGF control of glomerular filtration rate, in *The Juxtaglomerular Apparatus*, edited by AEG PERSSON and U BOBERG, Amsterdam, Elsevier Science Publishers, 1988, pp. 387–406
13. SCHNERMANN J, BRIGGS JP: Interaction between loop of Henle flow and arterial pressure as determinants of glomerular pressure. *Am J Physiol* 256 (Renal Fluid Electrol Physiol 25):F421–F429, 1989
14. SAKAI T, HALLMAN E, MARSH DJ: Frequency domain analysis of renal autoregulation in the rat. *Am J Physiol* 250 (Renal Fluid Electrol Physiol 19):F364–F373, 1986
15. CASELLAS D, NAVAR LG: In vitro perfusion of juxtamedullary nephrons in rats. *Am J Physiol* 246:(Renal Fluid Electrol Physiol 15:)F349–F358, 1984
16. COHEN HJ, MARSH DJ, KAYSER B: Autoregulation in vasa recta of the rat kidney. *Am J Physiol* 245:(Renal Fluid Electrol Physiol 14:)F32–F40, 1983
17. MASON J, KAIN H, WELSCH J, SCHNERMANN J: The early phase of experimental acute renal failure. V. Effect of furosemide. *Pflügers Arch* 392:125–133, 1981
18. CASELLAS D, TAUGNER R: Renin status of the afferent arteriole and ultrastructure of the juxtaglomerular apparatus in 'superficial' juxtamedullary nephrons from rats. *Renal Physiol* 9:348–356, 1986
19. KRIZ W, BACHMANN S: Pre- and postglomerular arterioles of the kidney. *J Cardiovasc Pharmacol* 7(suppl 3):S24–S30, 1985
20. TAUGNER R, HACKENTHAL E: *The Juxtaglomerular Apparatus*. Berlin, Springer–Verlag, 1989, pp. 33–40
21. NOBLING R, BÜHRLE CP: Cell to cell coupling at the afferent arteriole: A combined electrophysiological and morphological study. (abstract) *Kidney Int* 36:307, 1989
22. MOORE LC, RICH A: Mathematical model of ascending myogenic responses to tubuloglomerular feedback-induced changes in afferent vascular resistance. (abstract) *Kidney Int* 33:412, 1988